

### Environmental Research, Technology Demonstration and Conference Project

<b>ECF Project:</b>	ECF 2018-35
<b>Project Title:</b>	Developing a multiplex PCR assay for rapid and quantitative differentiation between <i>E. coli</i> and cryptic <i>Escherichia</i> in the aquatic environments
<b>Applicant:</b>	Professor Lau Chun Kwan, Stanley, Department of Ocean Science, The Hong Kong University of Science and Technology
<b>Total Approved Grant:</b>	\$495,000
<b>Duration:</b>	1/7/2019 to 31/12/2020
<b>Project Status/Remarks:</b>	Completed
<b>Project Scope:</b>	Currently, <i>E. coli</i> and cryptic <i>Escherichia</i> can only be differentiated through the costly and time-consuming multi-locus sequence typing method or a cumbersome five-reaction PCR assay. Neither of these methods is time- and cost-effective enough for routine water quality monitoring. The purpose of this proposed project is to develop a multiplex PCR assay, in both colony and quantitative PCR formats, for rapid and quantitative differentiation between cryptic <i>Escherichia</i> and <i>E. coli</i> in marine and freshwater samples.
<b>Summary of the Findings/Outcomes:</b>	Fecal pollution of water resources is a major cause of waterborne diseases and habitat deterioration. For decades, the fecal bacterium <i>E. coli</i> has been used as the principal indicator of pollution in water quality monitoring programs in Hong Kong and other places in the world. The membrane filtration method typically used for the determination of <i>E. coli</i> concentration in water samples is straightforward and incurs low capital and operational costs. However, the method can be confounded by the presence of cryptic <i>Escherichia</i> in water samples. Cryptic <i>Escherichia</i> is closely related to <i>E. coli</i> , but it is ubiquitously present in the environment without association with fecal pollution. Therefore, the false detection of cryptic <i>Escherichia</i> as <i>E. coli</i> may cause inconvenience to the public and increase government expenditure unnecessarily in trying to trace and mitigate a pollution source that may not even exist. In this project, a multiplex PCR assay was developed for fast and accurate differentiation between cryptic <i>Escherichia</i> and <i>E. coli</i> in water samples. The multiplex PCR assay was made available in two different formats, namely colony PCR and quantitative PCR that work efficiently in tandem with the membrane filtration method for more accurate monitoring of water quality.